

*Supplementary Material*

**Microbial diversity in a submarine hydrothermal chimney from the serpentinized system of the Prony Bay (New Caledonia) over a 6 years period.**

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Supplementary Table 3. Comparison of the PHF microbial community with other serpentinizing environments\*.

OTU reference clone**	OTU size	% sequences / year			Taxonomy	Blast hit	% Identity	% Coverage	Alignment length	Source
		2005	2010	2011						
PHF-HY5ArA02 (KJ149164)	80	83.3				clone_Ced_A01 (KC574884)	98.75	100	796	
PHF-13-A45-G13 (KJ149156)	50			54.9	<i>Methanosarcinales</i>	clone_Ced_A01 (KC574884)	99.5	100	799	The Cedars
PHF-2A-A13-I03 (KJ149144)	12		10.1			clone_Ced_A01 (KC574884)	98.5	100	799	
PHF-2HY5ArE02 (KJ149165)	15	15.6				clone_SGXU588 (FJ791633)	95.88	100	796	
PHF-2AarcF09 (KJ149145)	10		8.4			clone_SGXU588 (FJ791633)	96.5	100	800	
PHF-13-A03-K23 (KJ149159)	7			7.7	<i>Methanosarcinales</i>	clone_SGXU457 (FJ791612)	95.51	100	800	LCHF
PHF-15-A33-A09 (KJ149157)	1			1.1		clone_SGXU755 (FJ791575)	96.62	100	800	
PHF-13-A49-I13 (KJ149158)	1			1.1		clone_SGXU541 (FJ791597)	95.88	100	800	
PHF-2AarcE07 (KJ149148)	29		24.4			clone_LC1231a82 (AY505052)	97.12	100	799	
PHF-13-A10-O21 (KJ149154)	4			4.4		clone_LC1231a82 (AY505052)	97.62	100	799	
PHF-2AarcE06 (KJ149146)	3		2.5			clone_LC1231a82 (AY505052)	96.7	100	799	
PHF-2AarcE03 (KJ149147)	3		2.5			clone_LC1231a82 (AY505052)	99.0	100	799	
PHF-13-A02-M23 (KJ149153)	1			1.1	<i>Thaumarchaeota</i>	clone_LC1231a82 (AY505052)	95.2	100	799	LCHF
PHF-2A-A40-O09 (KJ149135)	5		4.2			clone_LC1231a80 (AY505051)	96.1	100	799	
PHF-15-A26-C07 (KJ149150)	2			2.2		clone_LC1231a80 (AY505051)	95.1	100	799	
PHF-2C-A39-M21 (KJ149141)	1		0.8			clone_LC1231a80 (AY505051)	98.5	100	799	
PHF-15-A23-M05 (KJ149151)	3			3.3		clone_LC1231a51 (AY505046)	98.6	100	799	
PHF-2CarcH11 (KJ149133)	6		5.0		<i>Thaumarchaeota</i>	clone_F160cmFL245 (JN002684)	95.9	100	797	Leka ophiolite complex
PHF-2AarcE02 (KJ149137)	3		2.5			clone_DSA_OTU_2 (EF414498)	96.4	100	799	Mariana forearc mud volcanos
PHF-15-A24-O05 (KJ149152)	1			1.1	<i>Thaumarchaeota</i>	clone_DSA_OTU_1 (EF414497)	96.7	100	798	
PHF-13-A40-C15 (KJ149163)	12			13.2	<i>Thermococcales</i>	<i>Thermococcus</i> sp.Tc-S-85 (AB095157)	98.5	100	802	Central Indian Ridge
PHF-2C-B47-N24 (KJ149191)	15		12.6			clone_Ced_B01 (KC574890)	97.1	100	854	
PHF-13-B5-J02 (KJ149246)	5			5.4	<i>Chloroflexi</i>	clone_Ced_B01 (KC574890)	97.5	100	855	The Cedars
PHF-2A-B10-D04 (KJ149230)	3		2.5			clone_Ced_B01 (KC574890)	96.8	100	787	
PHF-HY7BaG04 (KJ149167)	2	2.8				clone_Ced_B01 (KC574890)	96.7	100	787	

PHF-2A-B16-P04 (KJ149228)	3	2.5		clone_NS1B1_K15 (KC574967)	97.2	76.6	849	
PHF-2HY3BaC10 (KJ149172)	9	12.7		<i>Betaproteobacteria</i> clone_LC1524B-50 (DQ270636)	99.6	98.9	878	LCHF
PHF-13-B7-N02 (KJ149245)	1		1.1	<i>Betaproteobacteria</i> clone_F160cmL260 (JN002878)	95.1	78.3	886	Leka ophiolite complex
PHF-2CbacC03 (KJ149184)	1	0.8		<i>Betaproteobacteria</i> clone_lagoon3_O12 (KC574828)	99.2	92.5	798	The Cedars
PHF-13-B21-J06 (KJ149248)	8		8.7	clone_CVCloAm3Ph15 (AM778006)	97.4	90.4	908	
PHF-2HY7-BaG08 (KJ159206)	5	7.0		clone_CVCloAm2Ph102 (AM777947)	97.7	100	817	
PHF-15-B47-N24 (KJ159201)	4		4.3	<i>Firmicutes</i> clone_CVCloAm2Ph102 (AM777947)	98.1	100	827	CVA
PHF-2HY2BaA08 (KJ149176)	1	1.4		clone_CVCloAm3Ph15 (AM778006)	97.0	90.4	908	
PHF-15-B21-J18 (KJ149239)	1		1.1	clone_CVCloAm2Ph23 (AM777965)	97.6	100	782	
PHF-2C-B42-D24 (KJ149193)	1	0.8		clone_CVCloAm3Ph98 (AM778028)	95.5	100	863	
PHF-13-B3-F02 (KJ149247)	4		4.3	<i>Alphaproteobacteria</i> clone_F155cmContig9 (JN002777)	95.2	96	833	Leka ophiolite complex
PHF-2C-B2-D14 (KJ149202)	1	0.8		clone_F155cmContig9 (JN002777)	95.2	96	833	
PHF-15-B16-P16 (KJ149243)	2		2.2	<i>Alphaproteobacteria</i> clone_SGXT605 (FJ792016)	95.9	100	820	LCHF
PHF-2A-B9-B04 (KJ149213)	1	0.8		clone_SGXT398 (FJ79183)	96.0	100	823	
PHF-15-B28-H20 (KJ159192)	1		1.1	clone_SGYF714 (FJ792439)	97.5	84.1	811	
PHF-2AbacB08 (KJ149210)	1	0.8		<i>Gammaproteobacteria</i> clone_SGXT449 (FJ791881)	96.1	100	876	LCHF
PHF-2A-B48-P12 (KJ149216)	1	0.8		clone_SGXT626 (FJ792035)	95.1	100	892	
PHF-13-B11-F04 (KJ149254)	2		2.2	<i>Deinococcus-Thermus</i> clone_CVCloAm1Ph47 (AM777992)	98.7	100	776	CVA

\* Results of blastn searches : only hits > 95% identity with sequences from serpentinizing environments deposited in Genbank (accession number in the brackets) are shown. References for LCHF (Mid Atlantic Ridge): Schrenk et al., 2004; Brazelton et al., 2006; Brazelton et al., 2010; Gerasimchuk et al., 2010; Roussel et al., 2011; for The Cedars: Suzuki et al., 2013; for CVA: Tiago et al., 2004, Tiago and Veríssimo, 2013; for Kairei (Central Indian Ridge) : Takai et al., 2004; for Mariana Forearc: Curtis et al., 2012; for Rainbow (Mid Atlantic Ridge): Roussel et al., 2011; for Leka (Norway): Daae et al., 2013; for Del Puerto Ophiolite (California): Blank et al., 2009

\*\* In red, sequences from 2005 clone library, in green from 2010 and in blue from 2011.

- Blank, J., Green, S., Blake, D., Valley, J., Kita, N., Treiman, A., and Dobson, P. (2009). An alkaline spring system within the Del Puerto Ophiolite (California, USA): a Mars analog site. *Planet. Sp. Sci.* 57, 533-540.
- Brazelton, W.J., Schrenk, M.O., Kelley, D.S., and Baross, J.A. (2006). Methane- and sulfur-metabolizing microbial communities dominate the Lost City Hydrothermal Field Ecosystem. *Appl. Environ. Microb.* 72, 6257-6270. doi: 10.1128/aem.00574-06.
- Brazelton, W.J., Sogin, M.L., and Baross, J.A. (2010). Multiple scales of diversification within natural populations of archaea in hydrothermal chimney biofilms. *Environ. Microbiol. Rep.* 2, 236-242.
- Curtis, A.C., Wheat, C.G., Fryer, P., and Moyer, C.L. (2012). Mariana Forearc serpentinite mud volcanoes harbor novel communities of extremophilic Archaea. *Geomicrobiol. J.* 30, 430-441. doi: 10.1080/01490451.2012.705226.
- Daae, F.L., Økland, I., Dahle, H., Jørgensen, S.L., Thorseth, I.H., and Pedersen, R.B. (2013). Microbial life associated with low-temperature alteration of ultramafic rocks in the Leka ophiolite complex. *Geobiology* 11, 318-339.
- Gerasimchuk, A., Shatalov, A., Novikov, A., Butorova, O., Pimenov, N., Lein, A., Yanenko, A., and Karnachuk, O. (2010). The search for sulfate-reducing bacteria in mat samples from the lost city hydrothermal field by molecular cloning. *Microbiology* 79, 96-105.
- Roussel, E.G., Konn, C., Charlou, J.-L., Donval, J.-P., Fouquet, Y., Querellou, J., Prieur, D., and Cambon Bonavita, M.-A. (2011). Comparison of microbial communities associated with three Atlantic ultramafic hydrothermal systems. *FEMS Microb. Ecol.* 77, 647-665.
- Schrenk, M.O., Kelley, D.S., Bolton, S.A., and Baross, J.A. (2004). Low archaeal diversity linked to subseafloor geochemical processes at the Lost City Hydrothermal Field, Mid-Atlantic Ridge. *Environ. Microbiol.* 6, 1086-1095.
- Suzuki, S., Ishii, S.I., Wu, A., Cheung, A., Tenney, A., Wanger, G., Kuenen, J.G., and Nealson, K.H. (2013). Microbial diversity in The Cedars, an ultrabasic, ultrareducing, and low salinity serpentinizing ecosystem. *Proc. Natl. Acad. Sci. U.S.A.* doi: 10.1073/pnas.1302426110.
- Takai, K.H., Gamo, T., Tsunogai, U., Nakayama, N., Hirayama, H., Nealson, K.H., and Horikoshi, K. (2004). Geochemical and microbiological evidence for a hydrogen-based, hyperthermophilic subsurface lithoautotrophic microbial ecosystem (HyperSLiME) beneath an active deep-sea hydrothermal field. *Extremophiles* 8, 269 - 282.
- Tiago, I., Chung, A.P., and Verissimo, A. (2004). Bacterial diversity in a nonsaline alkaline environment: heterotrophic aerobic populations. *Appl. Environ. Microb.* 70, 7378-7387. doi: 10.1128/aem.70.12.7378-7387.2004.
- Tiago, I., and Verissimo, A. (2013). Microbial and functional diversity of a subterrestrial high pH groundwater associated to serpentinization. *Environ. Microbiol.* 15, 1687-1706.